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AN ORIGINAL PROBLEM IN AEROSPACE BIOLOGY: THE EFFECT OF MECHANICAL VIBRATIONS AT THE CELLULAR LEVEL

by P. Grognot, R. Loubière, and A. Pfister

Paper presented at the Second International Symposium on Basic Environmental Problems of Man in Space, Paris, June 14-18, 1965.

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Translation of "Un Problème Original en Biologie Aérospatiale:
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ABSTRACT

In mice vibrated for 7, 15, or 30 minutes, the authors find an anaphase change of about 40%. The sensitivity of mitosis to mechanical vibrations is confirmed.

Among the harmful factors which may be in operation during space flights, those which are most frequently encountered are acceleration, weightlessness, and ionizing radiation. The latter two factors can only be studied to the proper extent on board space vehicles. /1*

However, Arseneva, Antipov, and their collaborators had the merit to suspect a biological effect from mechanical vibrations at the cellular level of mammals, and to demonstrate this experimentally on the ground.

Using a vibrating table, these authors reproduced the characteristics of the vibrations encountered in a rocket. By subjecting mice to this action, they found that the proportion of anaphase malformations in hematopoietic marrow reached a maximum of 9.79% 24 hours after the experiment, while this proportion was only 2.61% in the control animals.

This resurgence of old mechanical theories has seemed to us to be of great interest from several points of view, and we have tried to discover this phenomenon in the cells of the neoplastic Ehrlich ascites tumor. We felt that as this tissue, which is rich in mitosis, increases at a fast rate, it would be particularly sensitive and would eventually make possible a macroscopic translation of the cytologic phenomena.

Material and Methods

We utilized a vibrating table having the following characteristics:

- Table with Goodmans excitor 8/600.

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*Note: Numbers in the margin indicate pagination in the original foreign text.

- The acceleration and amplitude were kept constant by utilizing a servo-system of the Rocher chronometer type.

- The constants were verified by means of a piezoelectric accelerometer, of the JP 09 SOPEMEA type.

This provided rectilinear, sinusoidal vibrations.

We retained the experimental data provided by the Russian authors:

- Frequency 70 cps;

- Amplitude 4/10 mm peak to peak.

In the first experimental series, we vibrated our animals for 15 minutes.

Subsequently, in order to determine the role played by time in the appearance of changes, we performed three experimental series which lasted 30, 7, and 4 minutes, respectively.

We employed male Swiss mice of pure lineage, all of which weighed more than 25 grams at the time of innoculation.

For the same experimental series, the control mice along with the others received 0.1 ml liquid of the ascites of the same donor mouse, which had been innoculated for six days.

The animals were weighed daily and were subjected to vibrations six days after the innoculation - i.e., in the phase of exponential growth of the tumor. They were killed 24 hours after the experiment, at the same time as the control animals.

However, some of the animals were saved and were weighed daily until /3 their natural death.

The ascites liquid, which was removed 24 hours after the experiment, was smeared on a plate and fixed with Carnoy liquid.

For each animal, two plates were colored by means of the usual methods for purposes of classification: MAY-GRUNWALD-GIEMSA and green of METHYL-PYRONINE.

The observation and the recording were performed by the same observer under fluorescence, with the aid of a LEITZ ORTHOLUX microscope having a mercury vapor lamp. The fluorochrome employed was acridine orange; it provided a green fluorescence for the ADN and a red fluorescence for the ARN.

In addition to observations on a dark background, which is extremely restful for the observer, this method makes it possible to discover most

remarkably the finest chromatidic filaments which are not always immediately visible on the usual colorations.

Results

Under these conditions, we counted the number of anaphases, and among them the number of malformed anaphases.

As the Russian authors found, we noted that these malformations consisted primarily of chromosome bridges. However, we found chromatidic fragments in about 20% of these anaphase malformations. These fragments were frequently united in the same cell with the bridges, but we have not separated them in the general table. In this table we indicate the total number of abnormal anaphases:

/4

	Number of Animals	Anaphases Calculated	Abnormal Anaphases	Percentage of Anomalies
Control Animals	20	1,520	207	13.6
Vibrations 30 Minutes	4	482	184	38.1
Vibrations 15 Minutes	8	605	264	43.6
Vibrations 7 Minutes	4	385	138	35.8
Vibrations 4 Minutes	4	309	63	15.4

This table shows us the following:

1. The proportion of changes in the anaphase is about three times greater in animals which were vibrated for 15 minutes than in the control animals.

2. This proportion varies very little, if the exposure time is doubled or if it is reduced by half.

3. On the other hand, four minutes of vibrations do not increase in a significant way this proportion of anaphase anomalies.

The threshold for the appearance of these changes therefore seems to be located between 4 and 7 minutes, for the type of vibration under consideration.

In the group of animals which died naturally, due to the neoplastic Ehrlich

ascites tumor, we did not discover any significant difference between the control animals and any of the animals which were vibrated, with respect to:

- duration of life, /5

- weight,

- any clinical indication whatever.

Comments

We therefore appear to have confirmed the sensitivity of mitosis to mechanical vibrations which may be encountered during space voyages.

Two questions may then be posed:

- What are the mechanisms leading to such changes?

- What are the possible consequences on the entire organism?

We shall avoid the first question, because we do not have sufficient information to answer it. We shall simply reply that several arguments have been advanced by the Russian authors which seem to show that the chromosome damages are different, whether they are produced by the ionizing radiation or other flight factors - including mechanical vibrations. The principal argument seems to be the smaller percentage of chromosome fragments in the vibrated animals. However, it seems premature to isolate the spindle changes as the primum movens of these damages.

Whatever may be the mechanism involved, we have obtained an anaphase change of about 40% in the animals vibrated for 7, 15, or 30 minutes. However, we have not been able to discover any change in the evolution of the neoplastic Ehrlich ascites tumor.

We therefore may conclude that these modifications have no ill effect on the growth of tumoral tissue during full development.

This seems even more surprising due to the percentage of changes encountered, and we wondered whether this phenomenon were not related to a particular method of growth for neoplastic tissue. /6

Although this study falls outside of the framework of space biology, we shall present a rapid summary, even though it may be only an illustration of the impulse which new research can give to the works of classical biology.

Starting with the hypothesis that the growth of the ascites tumor is not a simple multiplication of injected cells, but entails active participation of the host, we examined certain abdominal organs of the experimental animals 4, 12, 24, and 48 hours after injection of the cells.

We were thus able to objectify the active participation of the peritoneal serous membrane in the cellular proliferation, as well as the unusual modifications of the exocrine pancreas during the evolution of the ascites.

Therefore, the changes in the anaphase are not sufficient to restrain the tumoral evolution. It is possible that even in the case of cellular death, the residue of the cell (let us say, a virus) can induce the appearance of new neoplastic cells.

In our opinion, as well as in the opinion of the Russian authors, the problem of the effect of mechanical vibrations on a tissue during the course of proliferation still remains in its entirety. Genetics and embryological studies should make it possible to clarify this new problem posed by the conquest of space.

Report of the Center for Information and Research on Aeronautical Medicine (Director: General Practitioner Raboutet).

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